Physiology & Behavior 107 (2012) 781–786

Contents lists available at SciVerse ScienceDirect

Physiology & Behavior

journal homepage: www.elsevier.com/locate/phb

Versatility of the mouse reversal/set-shifting test: Effects of topiramate and sex

Gregory B. Bissonette [a,b], Michelle D. Lande [a], Gabriela J. Martins [a,b], Elizabeth M. Powell [a,b,c,*]

[a] Department of Anatomy & Neurobiology, University of Maryland School of Medicine, Baltimore, MD, 21201, USA
[b] Program in Neuroscience, University of Maryland, Baltimore, Baltimore, MD 21201, USA
[c] Department of Psychiatry, University of Maryland School of Medicine, Baltimore, MD, 21201, USA

ARTICLE INFO

Article history:
Received 21 December 2011
Received in revised form 19 May 2012
Accepted 21 May 2012

Keywords:
Reversal learning
Attentional set-shift
Topiramate
GABA
Female

ABSTRACT

The ability to learn a rule to guide behavior is crucial for cognition and executive function. However, in a constantly changing environment, flexibility in terms of learning and changing rules is paramount. Research suggests there may be common underlying causes for the similar rule learning impairments observed in many psychiatric disorders. One of these common anatomical manifestations involves deficits to the GABAergic system, particularly in the frontal cerebral cortical regions. Many common anti-epileptic drugs and mood stabilizers activate the GABA system with the reported adverse side effects of cognitive dysfunction. The mouse reversal/set-shifting test was used to evaluate effects in mice given topiramate, which is reported to impair attention in humans. Here we report that in mice topiramate prevents formation of the attentional set, but does not alter reversal learning. Differences in the GABA system are also found in many neuropsychiatric disorders that are more common in males, including schizophrenia and autism. Initial findings with the reversal/set-shifting task excluded female subjects. In this study, female mice tested on the standard reversal/set-shifting task showed similar reversal learning, but were not able to form the attentional set. The behavioral paradigm was modified and when presented with sufficient discrimination tasks, female mice performed the same as male mice, requiring the same number of trials to reach criterion and form the attentional set. The notable difference was that female mice had an extended latency to complete the trials for all discriminations. In summary, the reversal/set-shifting test can be used to screen for cognitive effects of potential therapeutic compounds in both male and female mice.

1. Introduction

Cognitive dysfunction is a common behavioral symptom of autism, Tourette syndrome, Rett syndrome and schizophrenia [1–3]. Patients that suffer frontal lobe deficiencies can easily learn and follow individual rules, but have great difficulty modifying their responses to new rules. Performance deficits are observed on the Wisconsin Card Sorting Test (WCST), in which the subject must sort a series of cards dependent upon changing rules, such as suit and color [4,5]. Patients can learn simple rules for sorting the cards, but they are unable to change established behavior once the relevant category changes [6–8]. Damage to the prefrontal cortex, specifically the dorsal lateral prefrontal cortex in humans, leads to impaired formation of the attentional set and shifting between sets, which is measured in the WCST by the intradimensional to extradimensional (ID–ED) shift [9–11]. In addition, patients with damage to orbitofrontal cortical areas are impaired in learning simple reversal tasks, in which the cues signaling correct and incorrect responses are switched [12].

The components of the WCST and reversal learning employed in patient studies have been modified and adapted for research animal models [13–16]. In agreement with the patient data, lesion studies in the dorsal lateral prefrontal cortex in the primate, and in medial prefrontal cortical in the rat, and most recently mouse, demonstrated that disruption in medial prefrontal areas reduces the ability to shift between attentional sets [13,14,17–21]. Lesion damage to the orbital frontal cortical (OFC) region or improper neural development of the OFC GABAergic interneurons impairs reversal learning [15,18,22–24].

The current study investigated the consequences of the anti-epileptic drug topiramate, which has been shown to increase GABA signaling [25–27]. Reversal learning and set-shifting are dependent upon the number of GABAergic interneurons in frontal cortical areas [24], implying a role for GABA. While many anti-epileptic drugs act to increase GABA levels and suppress seizure activity, the drugs may also impair cognition [28]. The anticonvulsant topiramate has known side effects of difficulty with concentration and memory problems in >10% of the patients [29,30]. In this study, the mouse reversal/set-shifting paradigm was applied to test if topiramate affects two types of flexible behavior, reversal learning and set-shifting, in mice, as it does in humans.

Many neuropsychiatric disorders present with select sex biases. For example, schizophrenia, autism and Tourette syndrome are more common in males than females [31–33]. The majority of rat and mouse studies using reversal and set-shifting tasks selectively used males

* Corresponding author at: Department of Anatomy & Neurobiology, University of Maryland School of Medicine, HSF II S251, 20 Penn Street, Baltimore, MD 21201, USA. Tel.: +1 410 706 8189; fax: +1 410 706 2512.
E-mail address: epowe001@umaryland.edu (E.M. Powell).

0031-9384/$ – see front matter © 2012 Elsevier Inc. All rights reserved.
doi:10.1016/j.physbeh.2012.05.018

[14,19,20,34–42] or grouped both sexes together [21]. To our knowledge, the effect of sex has not been examined. We evaluated whether both sexes performed similarly by testing female mice using the previously published test parameters [18], and then an altered paradigm with additional discrimination tasks.

The current results extend the knowledge beyond our previous lesion data [18]. The first experiment tests whether the anti-epileptic drug topiramate affects reversal learning or ID–ED, as suggested by the human reports. In the second experiment, we evaluate the responses of female mice on the reversal/set-shifting test. Finally, we modify the procedure to achieve the ID–ED shift and then compare the behavior of male and female mice.

2. Materials and methods

2.1. Animals

Adult C57BL/6 mice (>12 weeks of age) were purchased from the Jackson Laboratory (Bar Harbor, ME) and housed for 1–2 weeks prior to behavioral testing. All research procedures using mice were approved by the Institutional Animal Care and Use Committee at University of Maryland and conformed to NIH Guide for the Care and Use of Laboratory Animals. Mice were individually housed and food restricted to reach 85% baseline free-feeding weight with 12 h light cycle from 07:00 to 19:00 h with a room temperature of 20–22 °C and 40–50% relative humidity. To familiarize the mice with the testing materials, the materials were introduced into the home cage at least 1 day (d) prior. Food deprived mice were trained to perform compound discriminations by digging in bowls of scented media. Mice were tested in the late afternoon during the end of the light cycle.

2.2. Reversal/set-shift task

A set-shifting task was performed but modified from the task as described previously [18]. At the start of each trial, the mouse was placed in the testing arena to explore two bowls with combinations of odors and digging media until digging in one bowl to signify a choice. The bait was a piece of Honey Nut Cheerio cereal (~5 mg), and the cues, either olfactory (odor) or somatosensory and visual (texture of the digging medium which hides the bait) were altered and counterbalanced. All cues were presented in identical small animal food bowls (All Living Things Nibble Bowls, PetSmart) that were identical in color and size. Digging media was mixed with the odor (0.01% by volume) and Honey Nut Cheerio powder (0.1% by volume). All odors were ground dried spices (Penzeys, Milwaukee, WI; Hershey Chocolate Co, Hershey, PA; or McCormick Spice Co, Hunt Valley, MD) and unscented digging media was purchased from PetSmart (KayKob, bedding, wood chips, aquarium gravel, aquarium stone, kitty litter (2 types)) or local discount stores (cotton balls, feathers, moss, plastic pellets, shredded paper, perlite, bark, packing peanuts). The mice were housed in Softcard bedding. The digging media did not contain any components used in the animal bedding. On the first day of training, the mice were given 4 consecutive trials with the baited food bowl to ascertain that they could reliably dig. All mice were able to dig for the reward. The testing was performed over a 4 d period.

Mice were tested through a series of discriminations where the exemplar pair was changed, but the dimension (odor or medium) of the correct choice remained the same. The dimension was relevant if its attributes predicted outcome. For example, if odor was the relevant dimension, then the mouse was required to choose the correct odor from each pair and ignore the attributes of the digging medium. In this example, the digging medium is considered the irrelevant dimension.

The discriminations (Tables 1 and 3) were as follows:

- A single series of simple discriminations (SD) in which the mouse was presented with two choices of the relevant dimension and one choice of the irrelevant dimension (i.e. two odors within the same medium).
- A single series of compound discrimination (CD) in which the mouse was presented with the same choices of relevant dimension as in the SD and two choices of irrelevant dimensions (the exemplar used in the SD and a new exemplar).
- A series of multiple intradimensional shifts (IDS1, IDS2, IDS3,…) in which the mouse was presented with compound discriminations using two novel exemplars from the relevant and irrelevant dimensions for each IDS. The relevant dimension of the correct choice (i.e. odor) was maintained throughout the discriminations.
- An extradimensional shift (EDS) in which the mouse was presented with a novel compound discrimination, except for the first time the correct choice was an exemplar that was previously from the irrelevant dimension. The previously relevant dimension has become irrelevant.
- Reversal discriminations (CDrev, IDSrev, or EDSrev) in which the mouse was presented with the same set of exemplars as in the previous discrimination, but the stimulus-reward pairing was reversed within the relevant dimension.

The baited bowl was randomly presented on either side of the testing cage, and the relevant exemplar was randomly presented with the irrelevant exemplars. The trial was stopped if the mouse did not dig within 3 min in the testing cage. Stopped trials were uncommon (<3% of all trials), and they occurred most frequently during the SD. Aborted trials were not observed after completion of the CD and were not included in the latency calculations. The order of discriminations and exemplars was the same for all mice, but the direction of the extradimensional shift (EDS, odor to medium or medium to odor) was counterbalanced within each experimental group. A criterion of six consecutive correct trials was required to complete each

Table 1
Order of tasks for Experiment 1 — effect of topiramate.

Task	Dimension		Exemplar combinations	
	Relevant[a]	Irrelevant	Correct	Incorrect
SD	Odor (O)	Medium (M)	O1, M1	O2, M1
CD	Odor	Medium	O1, M1, M2	O2, M1, M2
CDrev	Odor	Medium	O2, M1, M2	O1, M1, M2
IDS1	Odor	Medium	O3, M3, M4	O4, M3, M4
IDS2	Odor	Medium	O5, M5, M6	O6, M5, M6
IDS3	Odor	Medium	O7, M7, M8	O8, M7, M8
IDS4	Odor	Medium	O9, M9, M10	O10, M9, M10
EDS	Medium	Odor	M11, O11, O12	M12, O11, O12
EDSrev	Medium	Odor	M12, O11, O12	M11, O11, O12

[a] The order of discriminations was the same for mice, but the relevant dimension, and thus the direction of the EDS (odor to medium or medium to odor) was counterbalanced within each experimental group. The number of trials required to reach criterion (6 correct consecutive trials) was independent of choice of relevant dimension.

Table 2
Exemplar combinations for Experiment 1 — effect of topiramate.

Pair	Exemplar	Dimension	
		Odor (O)	Medium (M)
1	1	Rosemary	Aspen bedding
	2	Cloves	Gravel
2	3	Cinnamon	Kaykob bedding
	4	Sage	Moss
3	5	Onion	Perlite
	6	Paprika	Bark
4	7	Garlic	Cat litter
	8	Coriander	Feathers
5	9	Thyme	Plastic pellets
	10	Black pepper	Cotton balls
6	11	Cumin	Shredded paper
	12	Cardamom	Packing peanut pieces